IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of:	**Stuart Newman**	
Application Serial No.:	**08/993,564**	**Examiner: D. Clark**
Filed:	**December 18, 1997**	**Art Unit: 1633**
For:	**Chimeric Embryos and Animals Containing Human Cells**	
Attorney Docket #:	**45010-00601**	

Box RESPONSES -- NO FEE
Assistant Commissioner for Trademarks
2900 Crystal Drive
Arlington, Virginia 22202-3513

DM Considered 4/12/01

Declaration of Martha Reed Herbert, M.D., Ph.D.

I, Martha Reed Herbert, declare as follows:

1. I have been employed by the Massachusetts General Hospital, Department: Pediatric Neurology for the past 7 ½ years. My business address is 149 13th Street Room 6012, Charlestown MA 02129. My home address is 31 Ames Street, Somerville MA 02145

2. The area of my concentration is pediatric neurology, and I have been working in this area for the past 6 ½ years.

3. My educational background is as follows: Undergraduate: BFA, 1972, California Institute of the Arts; Major field: Design. Ph.D., 1981, University of California, Santa Cruz, Board of Studies in History of Consciousness, concentration in philosophy of biology. M.D., Columbia University College of Physicians and Surgeons, 1991. Post-doctoral research

fellow at Boston University Center for the Philosophy and History of Science, 1981-82. Developmental Neuroimaging, Massachusetts General Hospital, 1995 to present.

4. Based upon my training and experience I am familiar with the subject matter of this patent application and am familiar with the level of knowledge of one of ordinary skill in the art, namely someone with a M.D. and/or a Ph.D. degree, with at least two years of post-doctoral training. The invention relates to the field of developmental biology; my own research is in a specialized subfield of this area, *viz.* brain development. My basic science training in pursuit of the M.D. degree, the basic science component of my training as an intern and resident in pediatrics and neurology, my fellowship in pediatric neurology, my training in molecular genetics at the Jackson Laboratories, Bar Harbor, ME, and my independent research in the field of brain development have provided me with an overview of animal and human embryology and equipped me to evaluate innovations in this field.

5. I have reviewed the U.S. Patent Application and the United States Patent and Trademark Office's ("PTO") Official Action rejecting the above-captioned application.

6. It is my understanding that the PTO has taken the position that the specification fails to provide an enabling disclosure for how to make and use the chimeric embryos of the invention. Furthermore, it is my understanding that the PTO has taken the position that the invention is obvious.

7. Through my training and experience in the fields of medicine and brain development I am aware of published literature that describes, in detail, specific technologies for creating intra- and interspecific embryo chimeras, establishing that the techniques are not only well known, but readily available, understood, and used by researchers of ordinary skill in the art for a wide variety of investigations. Nonetheless, the literature fails to disclose or even propose the making of a chimeric embryo, chimeric cell line, or chimeric animal that

employs a combination of human cells and the cells of a nonhuman primate or any other animal.

8. Based upon my training and experience it is my understanding that the methods for making intraspecific mammalian chimeras (e.g., tetraparental mice) have been disclosed in the literature since the 1960s. (Mintz, B., and Baker, W. W. (1967). Normal mammalian muscle differentiation and gene control of isocitrate dehydrogenase synthesis. *Proc Natl Acad Sci USA* **58**, 592-8). Additional publications describing the methods for making intraspecific mammalian chimeras include Gardner, R. L. (1968). Mouse chimeras obtained by the injection of cells into the blastocyst. *Nature* **220,** 596-7; Markert, C. L., and Petters, R. M. (1978). Manufactured hexaparental mice show that adults are derived from three embryonic cells. *Science* **202,** 56-8; Mystkowska, E. T., Ozdzenski, W., and Niemierko, A. (1979). Factors regulating the degree and extent of experimental chimaerism in the mouse. *J Embryol Exp Morphol* **51,** 217-2; Petters, R. M., and Markert, C. L. (1980). Production and reproductive performance of hexaparental and octaparental mice. *J Hered* **71,** 70-4; Landa, V. (1991). The construction of different types of aggregates and chimaeric aggregates from individual blastomeres of mouse 8-cell embryos. *Folia Biol (Praha)* **37,** 164-70. These methods are extremely versatile (note the production of intact organisms from even four separate embryos), and well known in the field of mammalian embryology.

9. Based upon my training and experience it is my understanding that methods for making interspecific mammalian chimeras (e.g., between mouse and rat or between two species of mouse), have been disclosed in the literature since the 1970's and 1980's, respectively. Interspecific chimeras were created using the techniques originally developed for intraspecific mammalian chimeras (the mixing of embryonic cells from two different species). (Stern, M. S. (1973) Chimaeras obtained by aggregation of mouse eggs with rat

eggs. *Nature* **243**, 472-3; Rossant, J., Croy, B. A., Chapman, V. M., Siracusa, L., and Clark, D. A. (1982) Interspecific chimeras in mammals: a new experimental system. *J Anim Sci* **55**, 1241-8). Techniques originally developed for the production of intraspecific mouse chimeras (Gardner, R. L. (1968) *op. cit.*) were versatile enough to be used to produce sheep-goat chimeric embryos. Techniques to produce sheep-goat chimeras by embryo manipulation and the use of interspecific chimerism to allow successful interspecific embryo transplantation in sheep and goats are well known in the literature (Fehilly, C. B., Willadsen, S. M., and Tucker, E. M. (1984) Interspecific Chimaerism Between Sheep and Goat. *Nature* **307**, 634-6) and known and understood by those of ordinary skill in the art.

10. It was known to one of ordinary skill in the art that the technologies for producing chimeric mammalian embryos are relatively insensitive to variations in procedure or species origin of the cells. Thus, techniques developed for mouse embryo culture have proved useful for the culture of other mammalian embryos. Furthermore, techniques developed for producing mouse-mouse chimeras proved useful in production of rabbit-rabbit chimeras, mouse-rat chimeras, and sheep-goat chimeras.

11. It is my scientific opinion that one of ordinary skill in the art would not anticipate that use of a different mammalian species, such as the human, would require "undue experimentation" for the design of a protocol for producing chimeras. Rather, many available references reflect the utility of these techniques across species. (See, for example Hammer, R. E. (1998). Egg culture: the foundation. *Int J Dev Biol* **42,** 833-9).

12. Although there will undoubtedly be slight differences in technique and culture conditions involved in applying techniques of chimera production to another mammalian species, one of ordinary skill in the art would not consider these slight variations to constitute "undue experimentation."

13. In conjunction with routine consultation of the available literature, the present invention would be adequately enabled to one of ordinary skill in the art.

14. It is my understanding that the PTO has taken the position that the outcome of how to make and use chimeric embryos composed of cells from both human and non-human mammals is unpredictable and is therefore not entitled to patent protection. However, all biotechnological procedures inherently lead to unpredictable outcomes. Genetic and other uncontrolled biological variability of organisms necessarily make outcomes unpredictable. This unpredictability is well known to those of ordinary skill in the art. Because one of the proposed uses of the human/non-human chimera is as a research tool in developmental biology, the variability of outcome, which will provide insight into how embryo cells of human and nonhuman species may communicate, is one of the useful aspects of this invention.

15. It is my understanding that the PTO has taken the position that the creation of a human/non-human embryo would be obvious to one of ordinary skill in the art.

16. Although the techniques for creating intra- and interspecific chimeric embryos are known, this field of science is unpredictable, and I am unaware of any instance of this invention being made, taught, or suggested in the scientific literature. While the references set forth by the PTO fairly and correctly characterize what is known in the art, those references do not describe the human/non-human chimeric invention claimed in the above-referenced application. Thus the invention as a whole would not be obvious to one of ordinary skill in the art.

17. Despite numerous publications containing ample teachings of the techniques disclosed, I have seen no proposals in the scientific literature for producing a chimeric embryo using human and animal cells, or using such embryos as model systems for

developmental biological research, or producing an animal from such embryos, or using such animals as research models in research on brain function and cognition, or using such animals as sources of transplantable tissues and organs for medical therapy. No medical doctor of my acquaintance has referred to the potential use of such chimeric animals as sources of transplantable tissues, although there has been a great deal of discussion in the medical field of the potential and problems of transgenic animals as sources of transplantable tissues and organs. Moreover, although it is well recognized that there is no good animal model for studying the developmental aspects of cognitive dysfunction such as autism (Caviness, V. (2001) Research strategies in autism: a story with two sides. *Curr. Opin. Neurol.*, in press), the use of human-nonhuman primate chimeras, which would likely be highly suitable for these studies, has not been mentioned in the scientific literature of this field.

18. Despite the disclosure in the scientific literature of sheep-goat chimeras, the state of the art itself in combination with that disclosure would not motivate one of ordinary skill in the art to make aggregates including human cells. Just because the technology is available, it does not mean that it would have been obvious to do the experiments as described in the present invention.

19. Based upon these facts, and my training and experience, it is my opinion that the scientific literature would enable one of ordinary skill in the art to create the human/non-human embryos of the claimed invention, but would not render the creation of a human/non-human chimera obvious.

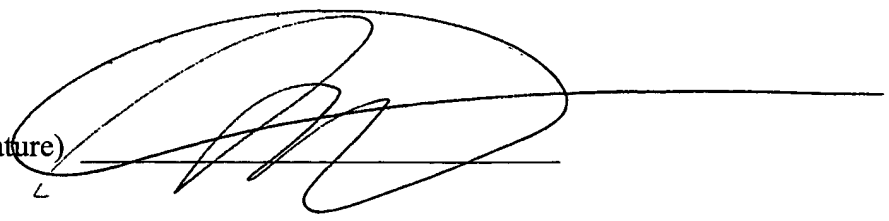
20. The assertion of the Examiner that xenografts, xenotransplants, or even allografts or allotransplants are chimeras, is not correct. For example, a baboon organ transplanted into a human does not result in a chimera. It is clear to me from the description of the invention

that the applicant is referring to what is known as "embryo chimeras." He is thus following the standard usage in the field. For example Gilbert, in the standard textbook *Developmental Biology* (Sinauer, 1997), defines "chimeric mice" as "the result of two or more early cleavage (usually 4- or 8-cell) embryos that have been artificially aggregated to form a composite embryo" (p. 187). Gilbert also refers to embryo chimeras being made from early stage embryo cells (blastomeres) and embryo stem (ES) cells. Although the term "chimera" has been used in other ways in the scientific literature, it would be clear to someone skilled in the art that the applicant is referring to chimerism achieved by using the cells of two or more early mammalian embryos, i.e., embryo chimeras.

21. In the production of embryo chimeras, because of uncertainties in the developmental process, the contribution each species will make to the chimera is not known in advance. Nonetheless, the definition of the resulting embryo and any organism that may develop from it as a chimera is not dependent on the knowledge before-hand of the ultimate physical and anatomical structure of the chimera.

I declare under penalty of perjury that the foregoing is true and correct to the best of my knowledge, information, and belief.

Executed on: Jan. 22, 2001



(Signature) _____

NAME (Printed) Martha R Herbert